

GCB Prelim Review

CAMB 550: *Caenorhabditis elegans*

23 April 2005

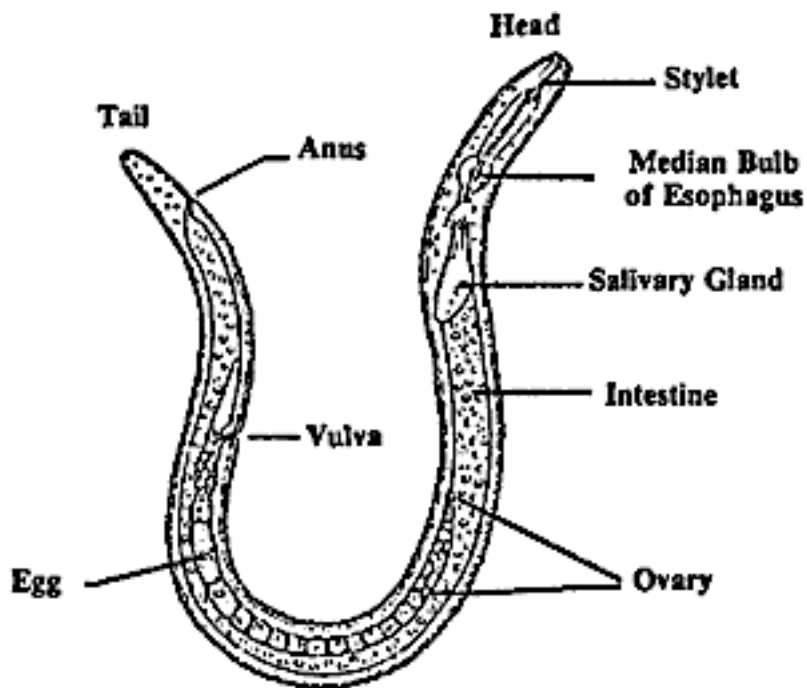
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Nomenclature

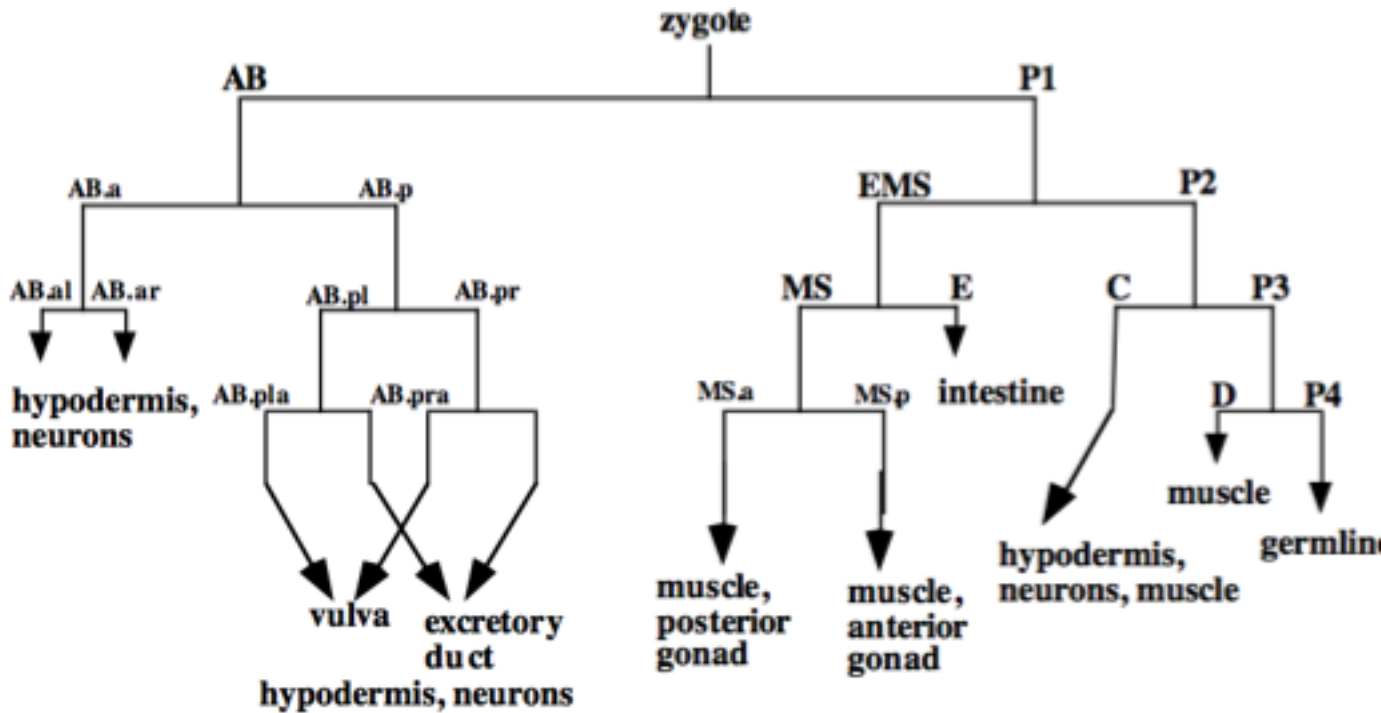
- *Caenorhabditis elegans*: Greek. kaino RECENT; rhabdos ROD; Latin elegans elegant)
- nematode (roundworm)
- genes: XXX-#(*allele*)

Anatomy

- Simple yet multicellular



Lineage



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- Invariant fate map (developmental patterning)
 - 959/1031 adult cells (hermaphrodite/male)
 - 302 neurons (neurobiology - sensory, neuromuscular)
 - 131 undergo apoptosis (apoptosis and signalling)
- Major fate patterns - differentiation through ancestry and cell-cell signaling
 - clonal - recapitulate ancestor's fate
 - stem-cell - one child cell retains ancestor fate, second differentiates

Features (cf other model organisms)

- Lifespan of 3 days
- Large broods 300-1000
 - amenable to forward genetic analysis
- Reverse genetic analysis faster (RNAi, morpholinos)
- Cheap & easy
- Cryogenics - freeze and revive
- Hermaphroditic
- Most of the same cell types, genetic pathways, etc as higher organisms

- Ablate cells using laser
- Transparent

Genetics

- diploid, 5 autosomes, 1 sex chromosome (X)
- 19,000 genes - relatively small (fly 13k)
- 100Mb (haploid) - (1/2x fly, 10x yeast, 30x human)
- Sex
 - Hermaphrodites (XX)
 - * make both sperm and eggs, mate with self, producing mostly hermaphrodites
 - Male (XO) infrequent .2% by meiotic non-disjunction
 - $XO \times XX \Rightarrow 1/2 XO + 1/2 XX$
- Good physical map at 50k resolution
- mutants: morphological (lethal, vulvaless), behavioral (dumpy, uncoordinated)

Reverse genetics

- Random chemical mutagenesis
- RNAi, but no targeted (ie homologous) gene knockouts
 - worms love RNA
 - microinjection - into adults - affects progeny
 - soaking - affects current worm
 - feeding - affects current plate of worms
 - Transgenes - under tissue or time specific promoter
 - * injection of DNA into germline (extrachromosomal array, multicopy)
 - insert array at various locations in the fate map and descendants will all carry the array
 - * “gene gun” bombardment (low copy integrants)
 - advantages
 - * quick and easy to get phenotypic information
 - * often get hypomorphic and knockout like effects
 - * multiple knockouts
 - disadvantages
 - * RNAi does not work for every gene
 - * possible cross-interference

Forward genetics (Positional cloning)

- linkage analysis, deficiency mapping to interval on genetic map
- genomics to predict location on physical map
- transgenics with clones to test for mutant rescue or use RNAi
- sequence coding region of mutant candidate gene(s), cf wild-type
- revise predictions using EST or cDNA clones

Signaling example: ras and vulvaless

- ancestor cell signals vulval precursors
 - ras necessary for 1° and 2° vulval fates (ras(-) => vulvaless)
 - ras sufficient for vulval fates (ras(gf) => multivulva)